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JUL 2 6 2002

Hogrete, H <110>

TECH CENTER 1600/2900

Hansen, Connie J

<120> Polymerase Enhancing Factor (PEF) Extracts, PEF Protein Complexes, Isolated PEF Proteins, and Methods for Purifying and Identifying

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<140> us 09/399,003

<141> 1999-09-20

<150> PCT/ US98/05497

<151> 1998-03-20

<150> US 08/957,709

<151> 1997-10-24

<150> US 08/822,774

1997-03-21

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33

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Thr Xaa His Xaa Cys Ser Thr Met Pro Gln Met Asn Leu Ser Asn Xaa 50 55 60

Asp Met Glu Arg Asp Leu Cys Arg Ser His Leu Xaa Gly Xaa Arg Val 65 70 75 80

Arg Gln Glu Thr Leu Thr Glu Glu Thr Ile Arg Gly Ala Gln Gly Xaa 85 90 95

Arg Phe Gln Arg Glu Arg Asn Ser Ser Val Phe Ser Ile Ala Ser Ser 100 105 110

Ile Ser Arg Val Lys Xaa Ser Met Xaa Ile Leu Ala Gly Trp Val Phe 115 120 125

Arg Asp Ser Asn Ser Xaa Asp Gly Pro Val Xaa Gln Lys Thr Ile Phe 130 135 140

Ala Ser Ser Ser Phe Ile Phe Leu Xaa Ile Lys Asn Pro Thr Ser Thr 145 150 155 160

Leu Val Pro Lys Asp Ile Val Cys Val Ile Thr Asn Lys Ile Leu Ala 165 170 175

Leu Phe Leu Ile Leu Tyr Ser Ile Leu Leu Ser Pro Ser Asn Leu Pro 180 185 190

Lys Ile Asn Leu Gly Ser Ile His Ser Leu Leu Ser Phe Lys Phe Leu 195 200 205

Xaa Ile Arg Thr Xaa Phe Arg Lys Met Ser Asn Ser Xaa Phe Pro Val 210 215 220

Lys Leu Thr Xaa Lys Ser Leu Xaa Xaa Xaa Phe Leu Xaa Phe Pro Lys Page 20 Pro Leu Ile Phe Pro Xaa 245

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Glu Gly Val Ile Gly Ser Phe Ala Trp Val Asp Pro Gly Trp Asp Gly 35 40 45

Asn Leu Thr Leu Met Leu Tyr Asn Ala Ser Asn Glu Pro Val Glu Leu 50 60

Arg Tyr Gly Glu Arg Phe Val Gln Ile Ala Phe Ile Arg Leu Glu Gly 65 70 75 80

Pro Ala Arg Asn Pro Tyr Arg Gly Asn Tyr Gln Gly Ser Thr Arg Leu 85 90 95

Ala Phe Ser Lys Arg Lys Lys Leu Xaa Arg Leu Phe Asn Ser Ile Leu 100 105 110

Asn Ile Ser Cys Glu Val Ile Asn Val Asn Thr Cys Trp Val Gly Phe 115 120 125

Xaa Gly Phe Lys Leu Val Arg Trp Ala Cys Ile Ala Glu Asn Tyr Phe 130 140

Cys Leu Phe Phe Ile Tyr Leu Ser Val Asn Lys Lys Ser Asn Ile His 145 150 155 160

Thr Ser Ser Lys Arg Tyr Cys Leu Arg Asp Tyr Gln Gln Asp Leu Gly
165 170 175

Ile Ile Phe Asp Leu Ile Leu Tyr Ser Pro Phe Ser Leu Gln Phe Ala 180 185 190

Gln Asn Lys Pro Gly Xaa Tyr Thr Phe Thr Pro Leu Phe Xaa Ile Pro 195 200 205

Ile Asn Ser Tyr Ile Val Xaa Lys Asn Val Lys Phe Phe Xaa Pro Cys 210 220

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Thr Pro Asn Phe Pro Leu 245

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Arg Arg Gly Tyr Trp Phe Phe Cys Leu Gly Xaa Pro Arg Met Gly Trp 35 40 45

Lys Leu Asn Thr Asn Ala Leu Gln Cys Leu Lys Xaa Thr Cys Arg Ile Page 22

60

Lys Ile Trp Arg Glu Ile Cys Ala Asp Arg Ile Tyr Lys Ala Arg Gly 65 70 75 80

Ser Gly Lys Lys Pro Leu Gln Arg Lys Leu Ser Gly Glu His Lys Val 85 90 95

Ser Val Phe Lys Glu Lys Glu Thr Leu Ala Ser Phe Gln Xaa His Pro 100 105 110

Gln Tyr Leu Val Xaa Ser Asn Gln Cys Lys Tyr Leu Leu Gly Gly Phe 115 120 125

Leu Gly Ile Gln Thr Arg Lys Met Gly Leu Tyr Ser Arg Lys Leu Phe 130 140

Leu Pro Leu Leu His Leu Ser Phe Cys Glu Xaa Lys Ile Gln His Pro 145 150 155 160

His Xaa Phe Gln Lys Ile Leu Phe Ala Xaa Leu Pro Thr Arg Ser Trp 165 170 175

His Tyr Phe Xaa Ser Tyr Thr Leu Phe Ser Phe Leu Pro Pro Ile Cys 180 185 190

Pro Lys Xaa Thr Trp Val Val Tyr Ile His Ser Ser Leu Leu Asn Ser 195 200 205

Tyr Lys Phe Val His Ser Leu Glu Lys Cys Gln Ile Leu Xaa Ser Leu 210 215 220

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Val Gly Lys Lys Ile Val Xaa Xaa Xaa Pro Gly Ser Ile Ala Ala 20 25 30

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Lys

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20

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<211> 437

<212> PRT

<213> Pyrococcus furiosus

<220>

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<222> (1)..(437)

<223> "X" represents any amino acid

<400> 51

Met Ile Ser Glu Ile Met His Pro Thr Lys Leu Leu Lys Gly Thr Lys 1 10 15

Ser Lys Leu Leu Glu Asn Lys Lys Ile Leu Val Ala Val Thr Ser Ser 20 25 30

Ile Ala Ala Ile Glu Thr Pro Lys Leu Met Arg Glu Leu Ile Arg His $35 \hspace{1cm} 40 \hspace{1cm} 45$

Gly Ala Glu Val Tyr Cys Ile Ile Thr Glu Glu Thr Lys Lys Ile Ile Page 25

60

50

Gly Lys Glu Ala Leu Lys Phe Gly Cys Gly Asn Glu Val Tyr Glu Glu 65 70 75 80 Ile Thr Gly Xaa Xaa Xaa Xaa Xaa Asp Ile Glu His Ile Leu Leu Tyr 85 90 95 Xaa Xaa Xaa Asn Glu Cys Asp Cys Leu Leu Ile Tyr Pro Ala Thr $100 \hspace{1.5cm} 105 \hspace{1.5cm} 110$ Ala Asn Ile Ile Ser Lys Ile Asn Leu Gly Ile Ala Asp Asn Ile Val 115 120 125 Asn Thr Thr Ala Leu Met Phe Phe Gly Asn Lys Pro Ile Phe Ile Val 130 135 140 Pro Ala Met His Glu Asn Met Phe Asn Xaa Xaa Ala Ile Lys Arg His 145 150 155 160 Ile Asp Lys Leu Lys Glu Lys Asp Lys Ile Tyr Ile Ile Ser Pro Lys 165 170 175 Phe Glu Glu Xaa Xaa Xaa Xaa Xaa Gly Lys Ala Lys Val Ala Asn 180 185 190 Ile Glu Asp Val Val Lys Ala Val Ile Glu Lys Ile Gly Asn Asn Leu 195 200 205 Lys Lys Glu Gly Asn Arg Val Leu Ile Leu Asn Gly Gly Thr Val Glu 210 215 220 Phe Ile Asp Lys Val Arg Val Ile Ser Asn Leu Ser Ser Gly Lys Met 225 230 235 240 Gly Val Ala Leu Ala Glu Ala Phe Cys Lys Glu Gly Phe Tyr Val Glu 245 250 255 Val Ile Thr Ala Met Gly Leu Glu Pro Pro Tyr Tyr Ile Lys Asn His 260 265 270 Lys Val Leu Thr Ala Lys Glu Met Leu Asn Lys Ala Ile Glu Xaa Xaa 275 280 285 Leu Xaa Ala Lys Asp Phe Asp Ile Ile Ile Ser Ser Ala Ala Ile Ser 290 295 300 Asp Phe Thr Val Glu Ser Xaa Phe Glu Gly Lys Leu Ser Ser Glu Glu

106

315

320

Glu Xaa Xaa Xaa Leu Ile Leu Lys Leu Lys Arg Xaa Asn Pro Lys 325 330 335

Val Leu Glu Glu Leu Arg Arg Ile Tyr Lys Asp Xaa Lys Val Ile Ile 340 345 350

Gly Phe Lys Ala Glu Tyr Asn Leu Asp Glu Lys Glu Leu Ile Asn Arg 355 360 365

Ala Lys Glu Arg Leu Asn Lys Tyr Asn Leu Asn Met Ile Ile Ala Asn $370 \hspace{1cm} 375 \hspace{1cm} 380$

Asp Leu Ser Lys Xaa Xaa His Tyr Phe Gly Asp Asp Tyr Ile Glu Val 385 390 395 400

Tyr Ile Ile Thr Lys Tyr Glu Val Glu Lys Ile Ser Gly Ser Lys Lys 405 410 415

Xaa Glu Ile Ser Glu Arg Ile Val Glu Lys Val Lys Leu Val Lys 420 425 430

Ser Xaa Xaa Xaa Xaa 435

<210> 52

305

<211> 444

<212> PRT

<213> Pyrococcus furiosus

<220>

<221> MISC_FEATURE

<222> (1)..(444)

<223> "X" represents any amino acid

<400> 52

Met Lys Ala Arg Gln Gln Lys Tyr Cys Asp Lys Ile Ala Asn Phe Trp
1 10 15

Cys His Pro Thr Gly Lys Ile Ile Met Ser Leu Ala Gly Lys Lys Ile 20 25 30

Val Leu Gly Val Ser Gly Gly Ile Ala Ala Tyr Lys Thr Pro Glu Leu 35 40 45

Val Arg Arg Leu Arg Asp Arg Gly Ala Asp Val Arg Val Ala Met Thr 50 60

Glu Ala Ala Lys Ala Phe Ile Thr Pro Leu Ser Leu Gln Ala Val Ser 65 70 75 80

Gly Tyr Pro Val Ser Asp Ser Leu Leu Asp Pro Ala Ala Glu Ala Ala 85 90 95

Met Gly His Ile Glu Leu Gly Xaa Xaa Xaa Xaa Lys Trp Ala Asp Leu 100 105 110

Val Ile Leu Ala Pro Ala Thr Ala Asp Leu Ile Ala Arg Val Ala Ala 115 120 125

Gly Met Ala Asn Asp Leu Val Ser Thr Ile Cys Leu Ala Thr Pro Xaa 130 135 140

Xaa Ala Pro Val Ala Val Leu Pro Ala Met Asn Gln Gln Met Tyr Arg 145 150 155 160

Ala Ala Ala Thr Gln His Asn Leu Glu Val Leu Ala Xaa Ser Arg Gly 165 170 175

Leu Leu Ile Trp Gly Pro Asp Ser Gly Ser Gln Ala Cys Gly Asp Ile 180 185 190

Gly Pro Gly Arg Xaa Xaa Asp Pro Leu Thr Ile Val Asp Met Ala Val 195 200 205

Ala His Phe Ser Pro Val Asn Asp Leu Lys His Leu Asn Ile Met Ile 210 215 220

Thr Ala Gly Pro Thr Arg Glu Pro Leu Asp Pro Val Arg Tyr Ile Ser 235 230 235

Asn His Ser Ser Gly Lys Met Gly Phe Ala Ile Ala Ala Ala Ala Ala 245 250 255

Arg Arg Gly Ala Asn Val Thr Leu Val Ser Gly Pro Val Ser Leu Pro 260 265 270

Thr Pro Pro Phe Val Lys Arg Val Asp Val Met Thr Ala Leu Glu Met 275 280 285

Glu Ala Ala Val Asn Xaa Xaa Ala Ser Val Gln Gln Asn Ile Phe 290 295 300

Ile Gly Cys Ala Ala Val Ala Asp Tyr Arg Ala Ala Thr Val Ala Pro 305 310 315

Glu Lys Ile Lys Lys Gln Ala Thr Gln Gly Asp Glu Leu Thr Ile Lys 325 330 335

Met Val Lys Xaa Asn Pro Asp Ile Val Ala Gly Val Ala Ala Leu Lys 340 345 350

Asp His Arg Pro Tyr Val Val Gly Phe Ala Ala Glu Thr Asn Asn Xaa 355 360 365

Xaa Xaa Xaa Val Glu Glu Tyr Ala Arg Gln Lys Arg Ile Arg Lys Asn 370 380

Leu Asp Leu Ile Cys Ala Asn Asp Val Ser Gln Pro Thr Gln Gly Phe 385 390 395 400

Asn Ser Asp Asn Asn Ala Leu His Leu Phe Trp Gln Asp Gly Asp Lys 405 410 415

Val Leu Pro Leu Glu Arg Lys Glu Leu Leu Gly Gln Leu Leu Leu Asp 420 425 430

Glu Ile Val Thr Arg Tyr Asp Glu Lys Asn Arg Arg
435
440

<210> 53

<211> 14

<212> PRT

<213> Pyrococcus furiosus

<220>

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<222> (1)..(14)

<223> "X" represents any amino acid

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Xaa Gly Xaa Xaa Asp Xaa Xaa Xaa Xaa Gly Xaa Xaa Xaa Xaa Page 29

10

<210> 54

1

<211> 14

<212> PRT

<213> Pyrococcus furiosus

<400> 54

Phe Ala Trp Val Asp Pro Gly Trp Asp Gly Asn Thr Leu Met $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 55

<211> 14

<212> PRT

<213> Pyrococcus furiosus

<400> 55

Ala Gly Trp Ile Asp Ala Gly Phe Lys Gly Lys Ile Thr Leu 1 10

<210> 56

<211> 14

<212> PRT

<213> Pyrococcus furiosus

<400> 56

Ser Ala Val His Asp Pro Gly Tyr Glu Gly Arg Pro Glu Tyr $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 57

<211> 14

<212> PRT

<213> Pyrococcus furiosus

<400> 57

Pro Thr Ile Val Asp Ala Gly Phe Glu Gly Gln Leu Thr Ile Page 30

1 5 10 <210> 58 <211> 14 <212> PRT <213> Pyrococcus furiosus <400> 58 Ala His Arg Ile Asp Pro Gly Trp Ser Gly Cys Ile Val Leu $1 \hspace{1cm} 5 \hspace{1cm} 10$ <210> 59 <211> 24 <212> DNA <213> Artificial Sequence <220> <223> synthetic <400> 59 24 gagttaaatg cctacactgt atct <210> 60 <211> 24 <212> DNA <213> Artificial Sequence <220> <223> synthetic <400> 60 24 caggactcag aagctgctat cgaa <210> 61 <211> 24 <212> DNA <213> Artificial Sequence

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gctgggagaa gacttcactg g

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<400> 66

His His Val Lys Leu Ile Tyr Ala 1 5

<210> 67

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<213> Pyrococcus furiosus

<400> 67

Lys Tyr Asp Ala Val Ile Met Ala 1 5

<210> 68

<211> 7

<212> PRT

<213> Pyrococcus furiosus

<400> 68

Glu Glu Asn Gln Val Val Leu 1 5

Page 33

21

<210> 69

<211> 8

<212> PRT

<213> Pyrococcus furiosus

<400> 69

Pro Asp Trp Lys Ile Arg Lys Glu 5

<210> 70

<211> 471

<212> DNA

<213> Pyrococcus furiosus

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	ıc cagcaggtta			-		120
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ctaaccctc	g agaggataaa	gttgcccgac	gatgttatgg	gggatatgaa	gataaggagc	240
agtttagca	a gagaaggggt	tattggttct	tttgcttggg	ttgacccagg	atgggatgga	300
aacttaaca	c taatgctcta	caatgcctca	aatgaacctg	tcgaattaag	atatggagag	360
agatttgtg	c agatcgcatt	tataaggcta	gagggtccgg	caagaaaccc	ttacagagga	420
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<210> 71

<211> 156

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<213> Pyrococcus furiosus

<400> 71

Met Leu Leu Pro Asp Trp Lys Ile Arg Lys Glu Ile Leu Ile Glu Pro $1 \hspace{1cm} 5 \hspace{1cm} 10 \hspace{1cm} 15$

Phe Ser Glu Glu Ser Leu Gln Pro Ala Gly Tyr Asp Leu Arg Val Gly 20 25 30

Arg Glu Ala Phe Val Lys Gly Lys Leu Ile Asp Val Glu Lys Glu Gly 35 40 45

Lys Val Val Ile Pro Pro Arg Glu Tyr Ala Leu Ile Leu Thr Leu Glu 50 60

Arg Ile Lys Leu Pro Asp Asp Val Met Gly Asp Met Lys Ile Arg Ser 65 70 75 80

Ser Leu Ala Arg Glu Gly Val Ile Gly Ser Phe Ala Trp Val Asp Pro 85 90 95

Gly Trp Asp Gly Asn Leu Thr Leu Met Leu Tyr Asn Ala Ser Asn Glu 100 105 110

Pro Val Glu Leu Arg Tyr Gly Glu Arg Phe Val Gln Ile Ala Phe Ile 115 120 125

Arg Leu Glu Gly Pro Ala Arg Asn Pro Tyr Arg Gly Asn Tyr Gln Gly 130 140

Ser Thr Arg Leu Ala Phe Ser Lys Arg Lys Leu 145 150 155

<210> 72

<211> 13

<212> PRT

<213> uridine-binding motif

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<222> (1)..(13)

<223> "X" represents any amino acid

<400> 72

Xaa Gly Xaa Xaa Asp Xaa Xaa Xaa Gly Xaa Xaa Xaa 1 10

<210> 73

<211> 14

<212> PRT

<213> Pyrococcus furiosus

<400> 73

Phe Ala Trp Val Asp Pro Gly Trp Asp Gly Asn Thr Leu Met $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 74

<211> 14

<212> PRT

<213> Methanococcus jannaschii

<400> 74

Ala Gly Trp Ile Asp Ala Gly Phe Lys Gly Lys Ile Thr Leu $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 75

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<213> Methanococcus jannaschii put.

<400> 75

Ser Ala Val His Asp Pro Gly Tyr Glu Gly Arg Pro Glu Tyr $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 76

<211> 14

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<213> D. sulf.

<400> 76

Pro Thr Ile Val Asp Ala Gly Phe Glu Gly Gln Leu Thr Ile 10

<210> 77

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<213> Escherichia coli
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<400> 77

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<210> 78

<211> 14

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<213> Escherichia coli

<400> 78

Val Gly Leu Ile Asp Ser Asp Tyr Gln Gly Gln Leu Met Ile $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 79

<211> 14

<212> PRT

<213> Yeast

<400> 79

Ala Gly Val Val Asp Arg Asp Tyr Thr Gly Glu Val Lys Val $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 80

<211> 14

<212> PRT

<213> Homo sapiens

<400> 80

Ala Gly Val Ile Asp Glu Asp Tyr Arg Gly Asn Val Gly Val $1 \hspace{1cm} 5 \hspace{1cm} 10$

<210> 81

<211> 14

<212> PRT

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